HBV

To: Gladys Kist and Susan Shultz

From: Harold Varmus

Re: Grant application for 1985 Hepatitis B Virus Meeting

A. The Challenge of Hepatitis B Viruses.

The serendipitous discovery of the viral agent of a widely recognized form of infectious hepatitis in man in the 1960's has stimulated the growth of a new field of biomedical research, one that joins traditional approaches to infectious disease with the techniques of modern molecular biology, in an effort to understand and control a major cause of human suffering. It has been frequently estimated that well over 200 million people are currently infected with the human hepatitis B virus (HBV). In many parts of the world, virtually everyone is infected at least transiently and the 5-20% who remain chronically infected are at high risk of inflammatory hepatic disease and primary hepatocellular cancer; in addition, they serve as a ready source of further infection. In parts of the world (particularly Africa and Southeast Asia) where rates of infection are particularly high, HBV-associated hepatic cancer is the most common form of fatal cancer. Even in the United States, where infection is less common, at least 0.1% of the population is composed of chronic carriers of HBV and life-time infection rates may be as high as 10%.

From the first identification of HBV, it has been appreciated that the agent has many novel properties that distinguish it structurally and functionally from the known animal viruses. Virus particles have an unusual structure and are vastly outnumbered in serum from infected individuals by incomplete particles composed principally of viral surface antigen (sAg) without nucleic acid. The interactions between virus and host are manifestly complex, leading sometimes to an immune response that appears to eliminate the virus and sometimes to extraordinarily persistant infection with very high titres of virus in the bloodstream; furthermore, the extent of pathological effect may vary from undetectable to profound loss of hepatic cells, hepatic cirrhosis, or liver cancer. From the early 1970's, efforts to describe HBV in molecular terms have provided further examples of its oddities: the genome is composed of a small circle of double stranded DNA in which neither strand is closed, the virus particles include a DNA polymerase activity, and protein is linked to one of the DNA strands.

Despite the obvious medical importance of HBV and its intriguing biochemical features, efforts to apply traditional strategies for the study of animal viruses to HBV have been frustrated by several problems. First, the virus has remained refractory to efforts to propagate it in cell culture. Second, HBV is strongly hepatotropic and highly species specific, confining studies of its growth largely to naturally infected human subjects and experimentally infected chimpanzees. Third, until a few years ago, there were no known HBV-related viruses found among experimentally-manipulable animals.

Remarkable advances have occurred during the past few years in the study of HBV, attributable in large part to the application of molecular cloning techniques and to the discovery of viruses closely related to HBV in a number of animals, including the Eastern woodchuck, the California ground squirrel, and

the domestic duck. Growth of DNA copies of the genomes of all of these viruses in bacteria has permitted nucleotide sequencing (revealing the extraordinary organization of viral coding domains) and some expression of viral gene products. The genomes of the four best studied hepatitis B viruses are now known to contain at least 3-4 open reading frames that overlap each other on a single strand and may be translated from more than one initiation codon in some frames. The incompletely described programs for transciption and translation of hepatitis B virus genes indicate that a surprisingly large amount of information is extracted from relatively small genomes (ca. 3200 nucleotide pairs, the shortest of the known animal virus genomes). Although a more complete understanding of the coding potential of these viruses will be required to probe interactions with their hosts in detail, it has already been possible to use the newly available information to design vaccines for human use, to raise antibodies for the identification of heretofore unseen viral proteins, and to gain insight into the mechanisms of tissue tropism and host range.

The pursuit of those hepatitis B viruses native to woodchucks, ground squirrels, and ducks has, in a few years, exposed the central features of the hepatitis B life cycle and generated opportunities for conducting sophisticated genetic and biochemical studies of the fascinating biological properties of these viruses. At the heart of these new discoveries is the finding that the hepatitis B viruses reproduce their DNA genomes through RNA intermediates, suggesting that the viruses use a replicative strategy that is a permuted version of the life cycle of retroviruses and that they encode an RNA-directed DNA polymerase ("reverse transcriptase"). Efforts to pursue these predications form a substantial part of current activity in the study of hepatitis B viruses. These efforts are now supported by recent evidence that molecularly cloned genomes of hepatitis B viruses can initiate infection when introduced directly into the livers of susceptible animals. Thus it is now possible to test mutant and recombinant genomes produced by in vitro methods for their ability to grow and produce disease in the animal, providing an important supplement to functional tests of portions of genomes by DNA transformation of cultured cells.

Finally, molecular techniques and precedents from the study of other kinds of tumor viruses have been used to explore the contribution made by HBV to hepatic cancer. Many tumors and tumor cell lines have been shown to carry integrated and usually rearranged copies of HBV DNA, and it is now possible to ask whether the viral DNA is active in the carcinogenic mechanism.

B. Rational for a regular meeting on the molecular properties of hepatitis B viruses.

The progress in hepatitis B research briefly outlined in the preceding section has been accompanied by an influx of people from various fields - animal virologists, genetic engineers, biochemists, immunologists, pathologists - who now share interests in the molecular biology of hepatitis B viruses. Nevertheless, despite the rapid growth of the field of hepatitis B viruses, there has been no appropriate forum for serious scientific discussion among its members. Traditionally, hepatitis B viruses, especially HBV, have been included mainly in medically-oriented gatherings and generally excluded from (or ignored at) meetings addressed to tumor viruses or animal viruses. The only consistent forum has been the tri-annual hepatitis meetings, sprawling events attended by many hundreds of clinicians, pathologists, and blood bank experts, as well as virolo-

gists, and not conducive to the kind of concentrated attention upon the molecular biology of hepatitis B viruses that is now called for. This state of relative disorder is reinforced by an examination of the vehicles for publication of work in this area of research. People doing rather closely related experiments often publish in journals that reflect their academic backgrounds (e.g. Gastroenterology or Hepatology for those with clinical training, versus J. Virology, Cell, or Proceedings of the National Academy of Sciences for those with backgrounds in basic science). Furthermore, the review articles about the molecular aspects of these viruses are few and generally focussed rather narrowly. All of these factors support the idea that the time is ripe for a meeting of modest size and length that is specifically addressed to advances on the molecular level in hepatitis B research.

C. Composition of the meeting.

See notes from our get-together in May.

P.S. I have not supplied references. You could use as a general reference, Chapter 11 from Readings in Tumor Virology (CSH press). In fact, you might want to include a copy of that chapter's introduction, as I may have suggested before.